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In This Issue

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Salinity drives fish evolution

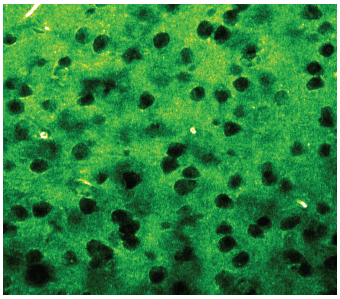
Image courtesy of Blackwell Publishing, Ltd.

The killifish *Fundulus heteroclitus*.

Atlantic killifish (*Fundulus heteroclitus*) are predominately saltwater fish distributed along steep salinity gradients across hundreds of miles of Atlantic coast estuaries. To determine whether salinity represents a barrier to gene flow across aquatic habitats in animals otherwise free to interbreed, Andrew Whitehead et al. (pp. 6193–6198) characterized the genetic structure of killifish distributed along the Potomac and James Rivers; regions where salinity gradients are in parallel as water flows into the salty Chesapeake Bay. The researchers captured fish from distinct salinity points in each river and found that killifish mitochondrial and nuclear gene markers matched at nearly identical salinities. Additional tests showed that killifish gill morphology changes at salinity levels around 0.5 ppt, and that gills in fish taken from water with less than 0.5 ppt adjusted well to being placed in fresh water, while fish taken from saltier areas downstream (1 ppt and greater) could not respond as quickly. The authors suggest that a threshold exists at salinities of 0.5–1.0 ppt, which triggers major genomic, cellular, physiological, and morphological adjustments in killifish gills, and that consequently, fish populations on either side of this threshold exhibit adaptations specific to their respective salt water or fresh water habitats. — J.M.

Live brain imaging without labels

Most high-resolution brain imaging techniques that help researchers visualize individual neurons in real time suffer from a shortfall: The techniques require the use of contrast-enhancing labels or fluorescent dyes, which could damage neurons or interfere with their function. Stefan Witte et al. (pp. 5970–5975) used an imaging technique called optical third-harmonic generation (THG) microscopy, which circumvents the need for labels, to visualize structures not only in brain slices but also in the brains of anesthetized mice.



Live neurons in mouse brain visualized with THG microscopy.

The technique exploits the specific geometry and lipid content of neurons to create a shadow-contrast image of brain structures in near-real-time. The authors report that THG microscopy enabled rapid, simultaneous imaging of neurons, white matter tracts, and blood vessels without significant damage to brain tissue or changes to neuronal function. Using the technique, the authors visualized individual neurons at depths greater than 300 μm in brain tissue, and reconstructed the neurons' approximate shapes and locations. In anesthetized mice, the authors obtained high-contrast images of neurons and blood vessels crisscrossing the brain at depths up to 200 μm, demonstrating the technique's ability to perform deep brain imaging in living animals. The authors suggest that THG microscopy could potentially prove useful in brain surgery for precisely guiding microscopic surgical tools and for performing minimally invasive, real-time, diagnostic brain tissue imaging. — P.N.

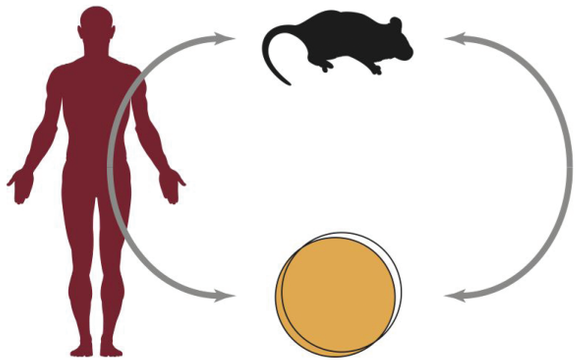
A genetic method to detect transplant rejection

To determine whether transplanted hearts might be rejected by the recipients, physicians rely on heart biopsies, which are risky and sometimes unreliable. Most alternative tests, which monitor the recipients' immune response, have poor predictive value. Thomas Snyder et al. (pp. 6229–6234)

attempted to diagnose the rejection of transplanted hearts by using a high throughput gene sequencing method that detects unique genetic signatures in donor-derived DNA circulating in the recipients' blood plasma. Elevated levels of circulating DNA from donor organs is a sign of cell death and, thus, of organ rejection. The authors quantified the relative amounts of donor-derived DNA in the plasma of transplant recipients and found that transplanted hearts were healthy in recipients in whom the average plasma level of donor DNA was below 1% of the total cell-free DNA. In patients in whom rejection was confirmed with a biopsy, the relative levels of donor DNA in the plasma hovered around 3%. When these patients received treatment for organ rejection, the levels returned to normal. Current genetic methods to monitor the health of transplanted organs typically rely on detecting the male sex chromosome in the plasma of female recipients of organs from male donors. But this group represents less than a quarter of all organ transplant recipients. Hence, the authors suggest, the sequencing method, if proven to be clinically useful, could help circumvent heart biopsies for detecting the rejection of transplanted organs. — P.N.

Re-creating communities of human gut bacteria

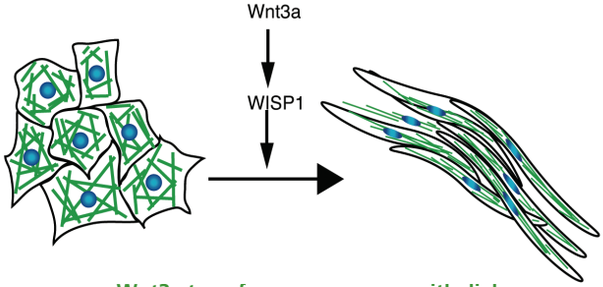
Researchers are increasingly recognizing the importance of gut bacteria to human health. To fully understand and exploit the human microbiome, researchers need to culture these bacteria, but exactly how closely cultured populations match the total community remains a mystery. Andrew Goodman et al. (pp. 6252–6257) cultured approximately 30,000 colonies of bacteria from the fecal samples of two unrelated humans, and compared the cultured bacteria's ribosomal RNA with bacterial RNA from the original samples. Nearly all of the ribosomal RNA sequences observed in the complete fecal samples shared the same phylum, class, and order as bacteria in the corresponding culture collections, and 56% of the sequences belonged to the same species-level phylogenetic type. To determine how cultured bacteria fared in vivo, the researchers transplanted cultured and complete bacterial communities into germ-free mice. Both community types exhibited similar colonization dynamics and distributions along the length of the gut, the study reports. Moreover, when mice were switched from a plant-based, low-fat diet to a high-fat, high-sugar "Western" diet, the transplanted culture collection responded similarly to the transplanted complete bacterial community. The study suggests that researchers can create, archive, transplant, and manipulate personalized human gut culture collections, which may help fuel the discovery of pre- or probiotics, and help clinicians treat nutritional deficiencies and other disorders. — J.M.



Populations of human gut bacteria remain stable in culture and in germ-free mice.

Small-molecule screening technique

Aberrant activation of the Wnt signaling pathway is linked to a variety of cancers, including those of the colon, liver, breast, and skin, typically due to faulty regulation of β-catenin responsive transcription (CRT). Inhibitors of CRT may prove effective therapies for cancer, but only if researchers can identify compounds that inhibit the role of β-catenin in transcription while preserving the protein's cytoskeletal function at cell–cell junctions. Foster Gonsalves et al. (pp. 5954–5963) developed a targeted screening technique, based on RNAi technology, that is capable of singling out compounds with greater selective activity against nuclear β-catenin. The authors assessed the effects of 14,977 compounds from a small-molecule library and identified several potential CRT inhibitors that were capable of blocking Wnt target genes in various mammalian and cancer cell lines. Furthermore, the CRT inhibitors specifically induced growth arrest in cells derived from human colon tumor biopsies and colon cancer cell lines with deregulated Wnt activity. The authors suggest that the technique could help identify small molecules targets for future anti-CRT cancer drugs, and further suggest that similar RNAi-based screening technologies could help target the activity of other signaling pathways implicated in human disease. — B.A.



Wnt3a transforms mammary epithelial cells into long, chord-like bundles.